Cell line designation: IPLB-Ekx4T

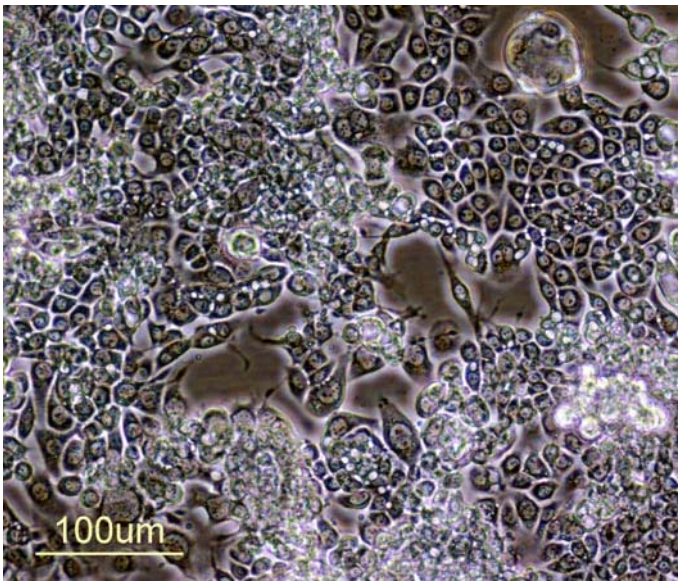
Tissue source: *Ephestia kuehniella*

Date initiated: September 2002

Morphology: fibroblastoid

Karyology: unknown

Culture medium: Ex-Cell 420 with 5% (v/v) heat inactivated fetal bovine serum (available from JRH Biosciences, Lenexa, KS)



Subculture procedure: At one week intervals, a confluent culture is split by trypsinization. Remove the medium from the culture and rinse the cells with buffer (see below)(use about 2 ml buffer to rinse a 25-cm² flask). The rinse is discarded and 1.0 ml trypsin (0.05 mg/ml VMF Trypsin, Worthington Biochem. cat. # LS04454 in buffer) is added. The flask is tilted to be sure entire monolayer is coated with solution then all but 0.3 ml is removed and discarded. Wait 5-10 min. Tap the culture on the bench top and look to see if some cells have detached. When they have, add 4.0 ml fresh medium and suspend cells by repeated pipetting over the flask surface. Transfer 0.7 ml cell suspension into 3.3 ml medium in a new flask (=~1:6 split). Cells are maintained at 26°C.

Buffer for trypsinization

NaCl	800 mg
KH_2PO_4	20 mg
KCl	20 mg
$Na_2HPO4{\times}7H_2O$	150 mg
Na_2EDTA	23 mg

in water to 100 ml

Adjust osmotic pressure with 15% NaCl to 350 mOsm/kg and pH to 7.0 with 2N NaOH. Filter sterilized through 0.2 µm filter and store at 4°C.

Comments: Cells are susceptible to baculoviruses originally isolated from *Autographa californica, Anagrapha falcifera, Heliothis armigera, Rachoplusia ou, Galleria mellonella* and *Plutella xylostella* while being only slightly susceptible to *Anticarsia gemmatalis* NPV, and with no apparent susceptibility to the NPVs from *Helicoverpa zea* or *Lymantria dispar*

Reference: Lynn, D. E., Ferkovich, F. M., 2004. **J. Insect Sci. 4** (9): 5pp. Available online: insectscience.org/4.9.

*Distribution of cell lines from the Insect Biocontrol Laboratory requires a letter on the recipient's letterhead agreeing that: (1) they will not be distributed to anyone outside your facility (2) they are for research purposes only, any commercial use must be agreed to in writing by the **Agricultural Research Service** and (3) publications describing research performed with the cells cites the appropriate reference and gives appropriate credit to the **Agricultural Research Service**.*